BioPharma research

Absolute Q Viral Titer Digital PCR Assays

Absolute quantification of viral vectors for cell and gene therapy development

As demand for cell and gene therapy continues to increase, so does demand for viral vectors and plasmid DNA development. Gene and cell therapies utilize a variety of viral vectors for gene transfer, including adeno-associated viruses (AAV), lentiviruses, retroviruses, herpes viruses, adenoviruses, and others. Among them, AAV, and its use in AAV-mediated gene therapy, is garnering the most interest due to the virus's safety profile—i.e., infection with the vector is not pathogenic and it cannot replicate on its own. The AAV viral vectors, as well as the messenger RNAs produced from plasmid DNA template, are the critical intermediates that produce and carry the chimeric antigen receptor (CAR) to T cells, enabling CAR-T therapies.

Accurately measuring viral titer for gene therapy development

Controlling the concentration of a potential therapeutic is difficult since viral particles are produced in living cells and then purified. The process of generating and purifying viral vectors is also susceptible to contamination by host cell DNA, mycoplasma, and other contaminants. The viral titer must be measured accurately, precisely, and consistently to ensure correct formulation. Harmful adventitious agents (virus, bacteria, and fungi) and contaminant DNA must be detected and screened out with the highest sensitivity to ensure product quality and safety. While qPCR remains the gold-standard tool for the quantitation of therapeutic viral vectors, such as AAV used in cell and gene therapies, digital PCR (dPCR) offers significant advantages over qPCR methods for the quantification of the target DNA molecules present in a sample. dPCR can provide an absolute count of nucleic acids, enabling the precise quantification of AAV vectors, bacterial contaminants, and residual host cell DNA. No reference standard is required, improving precision and removing a source of variation. dPCR is also less sensitive to contaminants that affect amplification, including those present in solutions used during the development of AAV-mediated gene therapies.

Applied Biosystems[™] Absolute Q[™] Viral Titer Digital PCR Assays enable easy, accurate, absolute quantification of viral vectors. The assays can be run individually, or as part of a custom multiplex assay that analyzes your target gene of interest. This enables you to measure the concentration and evaluate the quality of the viral vector for biopharma manufacturing and gene therapy research.

applied biosystems

Predesigned assays simplify your workflow

Predesigned Absolute Q Viral Titer Digital PCR Assays consist of a forward primer, a target-specific probe, and a reverse primer premixed together at specified concentrations. The assays require no further design, optimization, or verification. Just add your sample and reagent, then run your experiment.

- **Simple**—streamlined workflow for ease of use with your dPCR instrument
- **Fast**—Minimal hands-on time; results in 90 minutes when used with the QuantStudio Absolute Q Digital PCR System
- **Dependable**—analyze your data with confidence using verified assays backed by a performance guarantee*

We stand behind every predesigned Absolute Q assay you buy from us

We guarantee* the performance of all of our predesigned Absolute Q assays for dPCR experiments. Our application-specific portfolio of assays enables you to obtain the highest quality and performance available. These assays are designed and verified using up-to-date annotations and gold-standard Applied Biosystems[™] TaqMan[®] chemistry.

If an Absolute Q digital PCR assay does not perform according to conformance documentation, we will replace it at no cost or credit your account.*

Find out more at thermofisher.com/absoluteqassayguarantee

High precision in quantification across a 5-point AAV dilution series of AAV Viral particles using the Absolute Q Viral Titer dPCR Assay



Quantification of AAV viral vector dilution series using the Absolute Q Viral Titer dPCR Assay for AAV across 5-points across 5 Applied Biosystems^T QuantStudio^T Absolute Q^T MAP16 dPCR Plates demonstrating high linearity R2=0.995. Robust quantification observed with <4% CV and low plate to plate variation (Oneway ANOVA, Prob>f 0.1521; N=8 per plate).

Learn more about Absolute Q digital PCR assays at thermofisher.com/absoluteqassays

Ordering information

Target	Dye	Cat. No./ Assay ID
Absolute Q Viral Titer Digital PCR Assays		
Adeno-associated virus (AAV) ITR-2	VIC/MGB-NFQ	A53736/DTH49PU
Cytomegalovirus (CMV) promoter	VIC/QSY7	A52741
Custom dPCR assay		Please inquire

Powerfully simple digital PCR

Simplify your workflow even further by combining Absolute Q dPCR assays with the QuantStudio Absolute Q Digital PCR System—DNA sample to results can be done in <2 hours with minimal hands-on time. Moreover, there's no steep learning curve, as the workflow is identical to real-time PCR.

To complete your dPCR solution, use Applied Biosystems[™] Absolute Q[™] DNA Digital PCR Master Mix. Optimized for use with the QuantStudio Absolute Q Digital PCR System and Absolute Q assays, the 5X formulation enables analysis of a higher sample volume and delivers accurate quantification of DNA targets without using a standard curve.

Automate to accelerate

Reclaim valuable time and resources while helping to maximize throughput. The Applied Biosystems[™] QuantStudio[™] Absolute Q[™] AutoRun Digital PCR (dPCR) Suite is an integrated system solution that scales precise nucleic acid quantification while keeping it simple. With flexible instrument configurations and intelligent automation that supports up to 72 hours of hands-free operation, the AutoRun Suite helps to maximize highthroughput multiplex dPCR with a streamlined walk-away workflow.

The AutoRun Suite can be configured to integrate up to two QuantStudio Absolute Q dPCR instruments, and every Quantstudio Absolute Q dPCR System is compatible for an upgrade to the AutoRun Suite.

Applied Biosystems[™] QuantStudio[™] Absolute Q[™] dPCR solutions

QuantStudio Absolute Q Digital PCR System	-
	<u>nquire</u>
QuantStudio Absolute Q AutoRun dPCR Suite Please	<u>nquire</u>
Absolute Q DNA Digital PCR Master Mix (5X) A52490	
Absolute Q 1-step RT Digital PCR Master Mix (4X) A55146	

* Terms and conditions apply. To see full details of the guarantee, go to thermofisher.com/absoluteqassayguarantee

Experience powerfully simple dPCR with Absolute Q Viral Titer Digital PCR Assays at **thermofisher.com/dpcr-viraltiter**

For Research Use Only. Not for use in diagnostic procedures. © 2023 Thermo Fisher Scientific Inc. All rights reserved. All trademarks are the property of Thermo Fisher Scientific and its subsidiaries unless otherwise specified. QX ONE is a trademark of Bio-Rad Laboratories, Inc. QIAcuity is a trademark of Qiagen Group. TaqMan is a registered trademark of Roche Molecular Systems, Inc., used under permission and license. **EXT5279 0623**





applied biosystems